REMARKS

Claim 1-80 are pending, claims 1-17 have been elected (with SEQ ID NOS:36) and claims 18-80 have been withdrawn as drawn to non-elected subject matter.

Applicants herein cancel non-elected claims 18-80 without prejudice.

Applicant acknowledges the need to update the front page information to conform with the priority claims of record, and has done so.

Applicant acknowledges the Examiner's rejections of claims 1-17, under 35 U.S.C. § 112 ¶2, for indefiniteness. Applicant has responsively amended these claims to obviate this objection.

Applicant acknowledges the Examiner's rejections of claims 1-16, under 35 U.S.C. § 103(a), as being unpatentable over Duffy, etc. Applicant traverses this rejection, but has nonetheless made clarifying amendments to obviate this rejection.

Applicant acknowledges the Examiner's rejections of claims 1-13, under 35 U.S.C. § 103(a), as being unpatentable over Donini et al., etc. Applicant traverses this rejection, but has nonetheless made clarifying amendments to obviate this rejection.

No new matter has been added.

FORMALITIES

Claim objections. Applicant has amended claim 1 by substituting "a)"-"g)" for "a."-"g." to obviate this objection.

Conform Priority claim. Applicant has amended the front page, as described herein above, to conform with the priority claims of record.

Species election. Applicant has amended claim 17 in view of the provisional species election of record (*i.e.*, to SEQ ID NO:36), but maintains that recitation of all originally recited SEQ ID NOS is appropriate where independent claim 1 is determined to be allowable.

Rejections under 35 U.S.C. § 112 ¶2

The Examiner rejected claims 1-17 under 35 U.S.C. § 112 ¶2, for indefiniteness with respect to: a) recitation in claim 1 of "linker primer products" and "linker products" in view of lack of antecedent basis; b) lack of a detecting step in claim 1; and c) lack of antecedent basis for recitation of "nucleic acid fragments" in claim 3 (Office Action of 21 March 2005 at pages 3-4).

Applicant has responsively amended independent claim 1 to recite "linker primer fragments" and to additionally recite "to thereby detect the presence or absence of methylation of a CpG dinucleotide rich region" as suggested by the Examiner. Applicant has additionally amended claim 3 to recite "plurality of nucleic acid fragments" to provide proper antecedent basis.

Applicant, therefore, respectfully requests withdrawal of these rejections under 35 U.S.C. § 112 ¶2

Rejection under 35 U.S.C. § 103

Duffy:

The Examiner rejected claims 1-16, under 35 U.S.C. § 103(a), as being unpatentable over **Duffy** (U.S. Patent No. 5,871,917) in view of **Sapolsky** et al. (U.S. Patent No. 6,027,894), **Cross** et al. (Nature Genetics, 1994 6: 236-244) and **Pirrung** et al. (U.S. Patent No. 5,143,854) (Office Action of 21 March 2005 at pages 5-9).

The Examiner alleges that Duffy teaches (as claimed in applicant's claim 1) a process comprising: contacting a nucleic acid sequence with an enzyme (*e.g.*, MseI) which digests the nucleic acid sequence into fragments in which the CpG islands are preserved; attaching the fragments to linker primers to form linker primer products; contacting the linker primer products with a methylation-sensitive enzyme (*e.g.*, MspI) that digests fragments comprising *unmethylated* CpG sequences but not those comprising *methylated* CpG sequences to form a digestion product comprising methylated CpG island loci; amplifying the digestion product to form amplicons; labeling the amplicons; contacting the labeled amplicons with a screening array comprising a plurality of nucleic acid fragments on a solid support; and determining the presence or absence of labeled amplicons bound to the plurality of the nucleic acids affixed on the solid support. The

Examiner additionally states that the additional steps of Duffy are encompassed by Applicant's open claim language, and that while Duffy does not teach amplicon labeling or contacting amplicons with a screening array, Sapolsky et al. do, and that it would have been obvious to replace the Southern blotting of Duffy with the labeled amplification product detection on a screening array as taught by Sapolsky et al. for screening large numbers of sequence-specific genomic regions and to omits steps of Duffy for efficiency (Office Action of 21 March 2005 at pages 4-6).

Applicant respectfully traverses this rejection, because no *prima facie* case of obviousness can properly be made in view of the limited teachings of Duffy, alone or in combination.

Specifically, Duffy is based on, and teaches and requires the limitation that amplification is mediated by methylation-sensitive 'master' enzyme-generated ends; that is, in Duffy, amplification is either by means of primers that hybridize to the methylation-sensitive master enzyme-generated ends, or by means of amplifiable adaptors that are ligated to the methylation-sensitive master enzyme-generated ends. Moreover that recognition motif of the master enzyme is required to comprise a CpG dinucleotide sequence (see, *e.g.*, Duffy at: column 2, ll.54-67; column 3, ll. 30-33; column 4, ll. 8-10; column 9, ll. 23-28; column 10, ll., 36-39; etc., including the claims of Duffy).

By contrast, as argued by applicant, the present invention involves the pre-amplification cleavage of DNA into amplifiable-sized fragments using an enzyme that is not methylation-sensitive (*e.g.*, Mse I; TTAA; and preferably chosen not to contain any CpG sequences in the recognition motif), thus the amplification of the fragments (mediated through such DNA ends) is not mediated by methylation-sensitive master enzyme-generated ends (see, *e.g.*, the specification at: p.7, ll. 2-4; p. 25, ll. 10-12; p. 27, ll. 20-28; p. 28, ll. 1-7, etc.). The above-described limitation of Duffy has *profound* distinguishing consequences upon what sequences are represented in Duffy amplicons, which depend on methylation-sensitive master-end-dependent amplification. Significantly, many fragments that contain internal methylated CpG sequences will not be represented in Duffy amplicons by virtue of having a 'partner' enzyme-end, or if they don't, by virtue of being too large to be amplified. Moreover, in contrast to the present invention, the teachings of Duffy, which intentionally uses methylaion-sensitive enzymes (having a CpG dinucleotide in the recognition

motif thereof) to target CpG rich DNA regions, do not provide for amplicons from non-CpG-rich DNA regions.

Applicant, to further clarify this distinguishing aspect, has amended independent claim 1 to recite, in step a) "an enzyme that is not methylation-sensitive, lacks a CpG dinucleotide sequence in its recognition motif, and that cleaves the genomic DNA into fragments in which CpG islands are preserved and which have ends corresponding to the cleavage motif of the non-methylation-sensitive enzyme" and in step b) "ligating the fragments, via the ends corresponding to the cleavage motif of the non-methylation sensitive enzyme, to linker primers to form linker primer fragments."

Applicant has carefully considered, and thanks that Examiner for the analysis of claims 2-16 in view of Duffy, Cross and Sapolsky, but respectfully regard these additional assertions as *moot* in view of applicant's above arguments and responsive clarifying amendments.

Therefore, applicant respectfully requests withdrawal of the Examiner's 35 U.S.C. § 103(a) rejection with respect to claims 1-16, in view of amended claim 1, because the teachings and suggestions of Duffy (alone or in combination with Sapolsky et al., Cross et al., and/or Pirrung et al.) are entirely limited to the use methylation-sensitive master enzyme-generated ends to mediate amplification.

Additional Rejection under 35 U.S.C. § 103

Donini et al.:

The Examiner also rejected claims 1-13, under 35 U.S.C. § 103(a), as being unpatentable over **Donini** et al. (*Genome*, 1997, 40: 521-526), **Sapolsky** et al. (U.S. Patent No. 6,027,894), **Cross** et al. (*Nature Genetics*, 1994, 6: 236-244) and **Pirrung** et al. (U.S. Patent No. 5,143,854) (Office Action of 21 March 2005 at pages 9-13).

Specifically, the Examiner asserts that Donini et al teach a process for detecting the presence or absence of methylation of a CpG dinucleotide rich region of a nucleic acid sequence within a genome, the process comprising: contacting the nucleic acid sequence with an enzyme (*i.e.*, Mse) which digests the nucleic acid sequences into fragments in which CpG islands are preserved;

attaching the fragments to linker primers to form linker primer products; contacting the linder primer products with a methylation-sensitive enzyme (*i.e.*, Sse) which digests the linker products having unmethylated CpG dinucleotide sequences but no methylated CpG dinucleotide sequences to form a digestion product comprising methylated CpG island loci; amplifying the digestion product to form amplicons; labeling the amplicons; and determining the presence or absence of amplicons, wherein the labeled amplicons are run on a gel for determining the presence or absence of amplicons. The Examiner further states that additional steps of Donini are encompassed within applicant's open claim language, and while Donini do not teach contacting amplicons with a screening array, Sapolsky does, such that it would have been obvious to one of ordinary skill in the art to modify the Southern blotting of Donini with the screening array taught by Sapolsky, and to omit the additional amplification and ligation steps of Donini for efficiency.

Applicant respectfully traverses this rejection, because no *prima facie* case of obviousness can properly be made in view of the limited teachings of Donini et al, alone or in combination.

The instant method is not obvious in view of Donini (alone or in combination), because in the method of Donini, all of the fragments (*i.e.*, the entire complement of digested fragments) are amplified, because both Sse83871 and MseI linkers and primers are used (Donini et al., at page 522, under "Materials and Methods"). Thus, a mixture of amplified DNA products is produced, corresponding to all digestion products, including fragments corresponding to DNA regions that were methylated in the genomic DNA sample, and fragments corresponding to DNA regions that were not methylated in the genomic DNA sample.

Applicants have, in view of the Eaxminer's comments, clarified this distinguishing aspect by further amending independent claim 1 to recite "and wherein fragments cleaved by the methylation-sensitive enzyme are rendered non-amplifiable by the linker primers." However, even without such amendment, the argument presented herein above with respect to the teachings of Duffy are entirely applicable to the Examiner's Donini-based rejection. The amplification of Donini (alone or in combination) is limited to the requirement of mediation by at least one methylation-sensitive master enzyme-generated end (*i.e.*, Sse), and there is no disclosure, teaching or suggestion otherwise.

Applicant has carefully considered, and thanks that Examiner for the analysis of claims 2-13 in view of Duffy, Cross, Sapolsky and Pirrung, but respectfully regard these additional assertions as *moot* in view of applicant's above arguments and responsive clarifying amendments.

Applicant, therefore, respectfully requests withdrawal of the Examiner's 35 U.S.C. § 103(a)-based rejection of claims 1-13 in view of Donini et al., alone or in combination with the other asserted references.

Nonstatutory Double Patenting Rejection

The Examiner has rejected claims 1-17, under the judicially-created doctrine of non-statutory double patenting, as being unpatentable over claim 1-16 of applicant's U.S. Patent No. 6,605,432 (the '432 patent).

The Examiner asserts that the conflicting claimed subject matter, while not identical to, is nonetheless not patentably distinct from that claimed in the '432 patent; that is claims 1-16 and 21 of the'432 patent fall entirely within the scope of claims 1-17.

Applicants submit that the present application is commonly owned (ASSIGNEE: University of Missouri) with the allegedly conflicting '594 and '892 patents, and applicants are prepared to file a timely Terminal Disclaimer in compliance with 37 C.F.R. 3.73(b) upon the Examiner's indication of allowable subject matter.

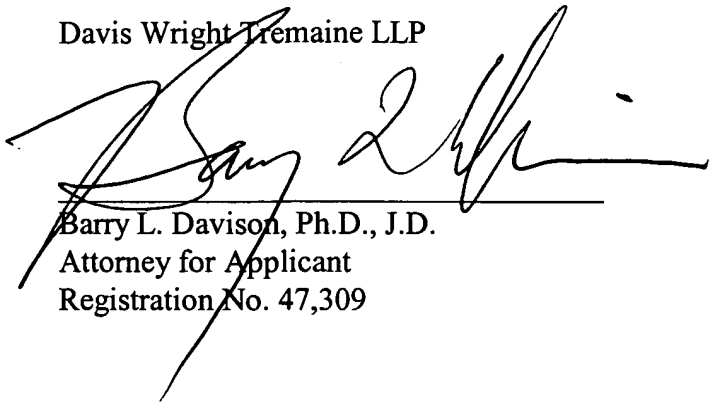
CONCLUSION

In view of the foregoing amendments and remarks, applicant respectfully requests entry of the present amendments, and allowance of all relevant claims as provided herein above. The Examiner is encouraged to phone applicant's attorney, Barry L. Davison, to resolve any outstanding issues.

No new matter has been added.

Respectfully submitted,

Davis Wright Tremaine LLP



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